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This was my first Research Paper - Frederick R. Miller, M.D., F.R.S.

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GALVANOTROPISM IN THE CRAYFISH.

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THE earliest observer of the phenomenon of galvanotropism was Mach[1], but the first detailed account was given by Hermann[2], who found that when free-swimming tadpoles or salmon-embryos were subjected to the influence of a galvanic current they came to rest with their heads towards the anode. Subsequently[3] he extended his earlier studies; the central nervous system was stimulated by an ascending or homodromic current, depressed or paralysed by a descending or antidromic current. Decapitated specimens also manifested the same peculiarities, *e.g.* unrest in ascending, rest in descending, currents. Even pieces of tail, provided they contained spinal cord, behaved in an entirely similar manner. In 1893 Blasius and Schweizer[4] carried out an extended series of observations on both vertebrates and invertebrates. The discovery of galvanotropism in the crayfish is due to these authors. They attempt to develop the view that a descending current causes anelectrotonus of the brain with consequent depression of reflexes, while an ascending current, on the other hand, leads to katelectrotonus of the brain with consequently increased reflex-irritability. Hermann and Matthias[5] criticise the position of Blasius and Schweizer. The brain is certainly not essential for the reaction, as one gets practically the same results in decapitated animals. Moreover it is impossible to determine the exact physiological electrodes, many of which undoubtedly exist within the central nervous system. More recently the subject has been approached by Loeb and his students, both as regards the higher and lower forms. Loeb and Maxwell[6], in an article on different crustaceans, state that

[1] Mach. *Bewegegungsempfindungen*, p. 53. Cited by Breuer, *Sitzungsb. der Wiener Akad.* CXIV. p. 27. 1905.

[2] *Pflüger's Arch.* XXXVII. p. 457. 1885.

[3] *Ibid.* XXXIX. p. 414. 1886.

[4] *Ibid.* LIII. p. 493. 1893.

[5] *Ibid.* LVII. p. 391. 1894.

[6] *Ibid.* LXIII. p. 121. 1896.

when medium-strength currents are used the above-mentioned stimulating and depressing influences are not observed. They explain the appearances by means of an ingenious though intricate theory.

In addition to the above work which has been carried out on the higher forms, much has also been done on the Protozoa; a great deal of this has been of a purely physiological character. Some observers, however, have attempted to give various physical explanations: for example, Carlgren[1] attempts to explain it by means of kataphoresis; Loeb and Budgett[2] by the action of the products of electrolysis, while lately Coehn and Barratt[3] have given an interpretation from the point of view of physical chemistry.

In view of the great interest attaching to the phenomenon it is strange that so comparatively little work has been done on the crayfish, which is rather unique among the higher forms in manifesting a galvanotropic attraction. I have therefore thought it desirable to make the following contribution to this subject.

Method. The animal used was the common Canadian crayfish, *Astacus fluviatilis.* The current employed was that installed in the laboratories; it is derived from the city-mains and is cut down by means of appropriate rheostats. For the most a current of 6 volts was used, but for some experiments it was increased to 15 volts. The fine gradations of current were secured by a rheostat, consisting of a bath filled with 10% $CuSO_4$ to which a few drops of H_2SO_4 were added: into this dipped two V-shaped copper electrodes, which could be moved vertically as well as horizontally through the solution and the desired strength of current in this way obtained. The experiments were carried out in a glass dish measuring $20 \times 5 \times 10$ cm. Like Blasius and Schweizer the writer made use of sheet-zinc electrodes, the cross-sectional area of which exposed to the water was 1750 sq. mm. For very large animals a larger bath was used, measuring $36 \times 10 \times 8$ cm., the area of the electrodes being 4800 sq. mm. The current was measured by means of a very accurate D'Arsonval galvanometer, which threw a beam of light upon a celluloid scale. The instrument was graduated so that 15 mm. deflection corresponded to a current through it of 0·00848 milliampères. Its resistance was 190 ohms. In the experiments it was shunted against a resistance of 0·1 ohm, and the current traversing the whole circuit determined. The currents were

[1] *Arch. f. Anat. u. Physiol.* p. 49. 1900.

[2] *Pflüger's Arch.* LXV. p. 518. 1897.

[3] *Verworn's Ztschr. f. d. allgem. Physiol.* V. p. 1. 1905.

estimated in Hermann's units, δ equals 1/1000 milliampère per sq. mm. of area exposed.

The crayfish when placed in a dish such as the above with zinc electrodes at opposite ends behaves as follows in response to the constant current. With a weak current the animal, if already facing the anode (antidromic position), moves forwards in the typical coordinated manner described by Bethe as normal for the crayfish[1]. If not in this position it turns to assume it and then moves forwards as before. With a strong current the animal in the antidromic position moves forwards very rapidly, but if the current is excessively strong the limbs become rigid and the animal is unable to advance. If a strong current is passed through an animal in the homodromic position it moves backwards by the powerful and characteristic tail-flexions: it does not in this instance first assume the antidromic position, but darts backwards to the anode. It is true the animal sometimes goes to the kathode, but it is undoubted that the main tendency is to move to the anode, where it invariably remains the longest and goes most readily. Such irregularities are probably due to the influence of the electrolytic products of dissociation.

When the head is towards the anode, at the moment of closure of the current, the telson is closed together, but soon spreads out again; the chela also undergoes a preliminary closure followed by opening: the abdomen is at the same time extended. When moving backwards to the anode the telson is spread out and the abdomen flexed underneath the thorax.

When using currents of moderate strength, one cannot observe any such definite orientations of the limbs as Loeb and Maxwell[2] describe for *Palæmonetes*. Only when the animals are exposed to strong currents is it possible to make out anything of this kind. The appearances of *Astacus* under these circumstances have been described and figured photographically by these authors. In the antidromic position one has the abdomen stretched out, the chelipeds flexed and the claws firmly closed. In the homodromic position the abdomen is flexed, the telson spread out, the antennæ are bent backwards, the antennules bent underneath the body, and the claws stretched open. Loeb and Maxwell had noticed that the muscle which opens the claw was here in a state of contraction. When the current is passed transversely through the animal one notices that the limbs are flexed on the side of the anode, extended on the side of the kathode, the claw on the anodic side is closed, that on the kathodic side is open.

[1] *Pflüger's Arch.* LXVIII. p. 456. 1897. [2] *loc. cit.*

It is doubtful whether Loeb and Maxwell, although describing these appearances, placed the correct interpretation upon them. The behaviour of the claw in the above instances really gives the key to the whole problem. When the head is towards the anode the claw is closed, when it is towards the kathode the claw is open; the appearances presented by all the muscles may be explained entirely by a reference to the well-known Ritter-Rollett phenomenon[1]. Ritter, at the beginning of the nineteenth century, studied the effect of stimulating the sciatic nerve in the frog, with the muscles of the limb intact. He found that with weaker stimuli one gets contraction of the flexor muscles, and consequently flexion of the limb, while with stronger stimuli the extensor muscles contract and marked extension is obtained. Rollett[2] in 1874 verified these earlier results with more suitable means and established the principle that antagonistic muscles, such as the flexors and extensors of a limb, possess, relatively to each other, different degrees of irritability. It has since been found that the principle involved in the Ritter-Rollett phenomenon is but part of a great general law. Thus Grützner[3] found that one strength of stimulation applied to the vagus caused narrowing of the glottis, while a stronger stimulus caused opening. Indeed it is very probable that wherever antagonistic groups of muscles exist these differences in irritability obtain. It is practically certain that this principle plays a very important *rôle* in the normal functioning of these different structures.

Probably the best and most interesting example of this phenomenon is afforded by the claw of the crayfish. Richet and Luchsinger[4] were the first to show that a weak stimulation of the nerve of the limb led to opening of the claw, while a stronger one caused closure. As is well known the opening and closing movements are carried out by means of two antagonistic muscles, a small abductor and a large adductor, the former contracting under the influence of a weaker, the latter of a stronger, stimulus. The question now is whether the result attained in any case is not merely the outcome of a struggle between the two muscles or whether there is such an arrangement that contraction in one muscle is associated with inhibition in its antagonist. This question

[1] A good account of this phenomenon is given by Osswald, *Pflüger's Arch.* L. p. 215. 1891.

[2] *Sitzungsb. d. Wiener Akad.* LXX. LXXI. LXXII. 1874–76. Cited by Biedermann, *Electrophysiologie.*

[3] *Breslauer ärtzl. Ztschr.* p. 190. 1883. Cited by Biedermann, *Electrophysiologie.*

[4] Richet. *Physiol. des muscles et des nerfs*, p. 274. 1882. Luchsinger, *Pflüger's Arch.* XXVIII. p. 60. 1882. Cited by Biedermann, *Electrophysiologie.*

has been answered by Piotrowski[1], who has studied the abducto adductor muscles separately; working in this way he found it possible to obtain genuine inhibitory effects by appropriate strengths of stimulation. It is particularly interesting in this connection to note that the strength of stimulation which produces contraction in one muscle produces inhibition in its antagonist. We have here obviously a remarkable peripheral mechanism, which enables the opposed muscles to act in absolute harmony relatively to each other.

The anatomical basis has been worked out very fully quite recently by Mangold[2]. He finds that each of the claw muscles is supplied by two sets of nerve fibres: not only so, but all the body muscles studied have been found to possess a double innervation. It is but natural then to assume that one set of fibres conducts motor, the other inhibitory, impulses. Inhibition by stimulation of peripheral nerves has been observed among other Invertebrates. For instance Pawlow[3] found the muscles which close the shells of molluscs to be supplied with motor and inhibitory fibres.

Effects somewhat analogous to the above are to be noticed in the Vertebrates. Thus if the flexor-reflex at the knee-joint is induced by stimulation of the foot the extensor muscles are found to be inhibited[4]. The inhibition necessary for the coordination of the antagonistic muscles of the crayfish claw is seen to be a peripheral phenomenon. Among the Vertebrates, on the other hand, it is associated with the intervention of the gray matter of the central nervous system[5]. The cause of this phenomenon in the crayfish is not at all certain. It may be, however, that the so-called "interference phenomenon" in nerves plays an important *rôle*[6].

Who can doubt now that the behaviour of the crayfish, when under the influence of a constant current, is not associated with such fundamental peculiarities of its neuro-musculature as these? When the head of the crayfish is towards the anode the claw is closed, when towards the kathode it is open. In the first case we must suppose the stimulus passing down the peripheral nerves to be of such a character that contraction is induced in one set of muscles, and inhibition in their

[1] This *Journal*, XIV. p. 163. 1893.
[2] *Verworn's Ztschr. f. d. allgem. Physiol.* V. p. 135. 1905.
[3] *Pflüger's Arch.* XXXVII. p. 6. 1885.
[4] Sherrington. *The Integrative Action of the Nervous System*, p. 88. 1906.
[5] *Ibid.* p. 85.
[6] Fuld. *Pflüger's Arch.* LXXXI. p. 381. 1900.

antagonists. As far as I am aware inhibition has not been demonstrated in the other body muscles, yet it undoubtedly occurs, for the anatomical basis is present, as mentioned above. We may be quite certain then that not only the appearances in the claw depend on these facts, but also the entire orientation of the animal. When the crayfish is in the antidromic position the current acts on the central nervous system in such a way that the impulses originating are of a strength suited to cause the contraction of one group of muscles, those of the limbs, body and telson with inhibition of the antagonistic group; when the animal is in the homodromic position the other group of muscles is stimulated, while the first group is now inhibited.

Biedermann[1] and all other investigators agree that the central nervous system is the chief point of action of the galvanic current. It was not inconceivable that reflex stimulation occurred from the sensory endings in the skin, but this possibility has been recently excluded by Breuer[2]. It would seem probable that the peripheral nerves must also receive stimulation, and this may be readily shown by the following simple experiment. The cheliped is cut off and placed in a dish of water, through which a current is then passed. It will be observed that the muscles are differently affected when the current is passed in opposite directions. That the appearances are due to the action of the current on the nerve can be readily shown, for if two holes are made in one of the proximal segments and platinum electrodes inserted, through which currents are passed, one obtains similar results. Studies of this kind have also been carried on by Biedermann[3], who finds that in general the closing and opening muscles behave in an exactly opposite manner as regards the constant current. Here then we have another undoubted factor, though a secondary one, in producing the orientations of the crayfish. Since the action of the current on the peripheral nerves is mainly manifested at the make and break, the tonic condition of the muscles producing the orientations of the intact animal must be due to the galvanisation of the nerve-cells of the ganglionic chain. Breuer[4] has lately arrived at a similar conclusion in regard to fish.

The question now might be raised: is the stimulation which undoubtedly occurs in the central nervous system, mainly of the nerve-fibres or of the nerve-cells? The answer is that undoubtedly both are

[1] *Ergeb. d. Physiol.* I. p. 195. 1902.
[2] *Sitzungsb. d. Wiener Akad.* CXIV. p. 45. 1905.
[3] *Electrophysiologie*, p. 601. 1895.
[4] *Sitzungsb. d. Wiener Akad.* CXIV. p. 47. 1905.

stimulated: the nerve-fibres which pass in a general antero-posterior direction must be stimulated by the movement of ions and colloids just as we have shown the peripheral nerves to be stimulated. The nerve-cells are also unquestionably stimulated, though probably not in the peculiar manner explained by Loeb and Maxwell. They had assumed that when the current is passed through the animal in one direction the cell-bodies of the neurones supplying one set of muscles are in katelectrotonus, while those of their antagonists are in anelectrotonus. They also supposed that one set of nerve-elements represent a crossed relationship to the muscles which they supply. It is true that anatomically certain neurones are seen, the axones of which cross each other in the middle line[1], but the ultimate distribution of these is notoriously uncertain. As far as the assumption of kat- and anelectrotonus is concerned it is a pure hypothesis, regarding which Biedermann expresses himself as rather sceptical[2]. Indeed Loeb[3], himself, in a subsequent communication on galvanotropism, finds that the behaviour of the skin-glands of Amblystoma, when this animal is exposed to a constant current, can only be interpreted by supposing the central nervous system to act as a uniform whole. He attempts to justify his theory in regard to crustaceans by stating that the nervous system in different groups of animals may behave differently, an assumption which Biedermann considers would not meet with very general agreement[4].

Briefly then the orientations which are observed can be explained as follows: when the animal is subjected to the influence of the current in one direction the central nervous system is stimulated and sends out impulses of such a strength that a certain group of muscles, possessing adequate irritabilities, is caused to contract, their antagonists being all inhibited; when the current is reversed the conditions are precisely the opposite. This stimulation occurring in the central nervous system is partly of the nerve-cells and partly of the nerve-fibres. Combining with this principle the action of the current on the peripheral nerves the orientations are readily explained. It is also clear from the above that the central nervous system of the crayfish is acted upon as a uniform whole and thus proves no exception to the general rule

[1] Retzius. *Biol. Untersuch.* I. p. 1. 1890. Also Allen, *Quart. Journ. Mic. Science*, XXXVI. p. 461. 1894.

[2] *Ergeb. d. Physiol.* I. p. 192. 1902.

[3] *Pflüger's Arch.* LXV. p. 308. 1897.

[4] *Ergeb. d. Physiol.* I. p. 194. 1902.

established for other forms. I shall refer subsequently to the real bearing, if any, of these orientations on galvanotropism.

As mentioned above Hermann[1] and Blasius and Schweizer[2] noticed that animals in the antidromic position were quiet, those in the homodromic were excited. They found this to apply in the fish, amphibian and crayfish. This is supported by the work of numerous writers; Blasius and Schweizer[3], working with the hind portions of frogs, obtained excitation in the ascending, and depression in the descending, current. The convulsions of strychninized frogs were inhibited by descending, and augmented by ascending, currents[4]. Uspensky[5] has shown that this is due to a specific action on the central nervous system: by means of needles he passed currents into the spinal cord: ascending currents, under these conditions, caused acceleration of the respirations, while descending currents led to their cessation. Legros and Onimus also arrived at similar results[6]. Biedermann[7], likewise, admits the truth of this general proposition.

Loeb and Maxwell have stated that by using currents of moderate strength the effects of excitation and depression are not observed. That they really do obtain, however, in the crayfish, as in the other forms studied, can be shown as follows. The animal is tied upon its back so that the appendages may be more readily observed. During the passage of a descending current the appendages are at rest, while during the passage of an ascending current they are in a condition of marked activity. It is true that when very strong currents are turned on in either direction one notices only orientations, but these do not appear with moderate currents which give rise to excitation and depression. Other important facts make it still more certain that this law applies in the crayfish. In the course of this work I have found that when any part of the ventral ganglionic chain is stimulated, as by the interrupted faradic current, the anal sphincter carries out rhythmical contractions. In a subsequent communication I shall report more fully on this phenomenon, which seems to afford an interesting analogy with the heart of Limulus, as described by Carlson[8].

[1] *loc. cit.*
[2] *loc. cit.*
[3] *loc. cit.* p. 528.
[4] *loc. cit.* 529.
[5] *Centralb. f. d. med. Wissensch.* VII. p. 577. 1869. Cited by Blasius and Schweizer.
[6] *Traité d'électricité médicale*, Paris, 1887. "Le courant ascendant excite la moelle et augmente les actions réflexes, tandisque le courant descendant empêche les actions réflexes et diminue l'excitabilité de la moelle." Cited by Blasius and Schweizer.
[7] *Ergeb. d. Physiol.* I. p. 189. 1902.
[8] *Amer. Journ. Physiol.* XII. p. 67. 1904.

If then the anal sphincter is active we have an indication that impulses are being sent out by the ventral chain of ganglia, that is to say, the ganglia are in a condition of increased irritability. One finds now that when a constant current in the ascending direction is passed through the animal the sphincter becomes active, while when a descending current is passed it is unaffected: this can only mean that in an ascending current the ganglia are in a condition of increased irritability, or the ascending current is stimulating. It is also clear that it is the anodic region itself whose irritability is increased. Blasius and Schweizer were of opinion that the anodic region was depressed in irritability while the kathodic region had its irritability increased. We see from the last-mentioned results, however, that exactly the opposite is the case, while it is at the same time clear that the ordinary results of electrotonus do not apply to the central nervous system.

That the action of the current in the above instances is a direct one on the central nervous system can be readily shown as follows. If one sections the cord between the thorax and the abdomen, thus leaving the abdominal ganglia intact, the sphincter first performs a series of rhythmical contractions owing to the stimulus of the cut. After the sphincter has come to rest one again finds that when the anode is towards the tail rhythmical activity is elicited, but not when the kathode is towards the tail. If the abdominal ganglia are now successively removed until only the sixth or last is left the typical action of the current is still manifested, *e.g.* activity when the anode is behind and rest when the kathode is behind. When, however, the last ganglion is removed all activity in response to the constant current is at once lost. Identical phenomena can be elicited by the application to the cord of platinum electrodes, through which a current is passed: even when the last ganglion is alone intact and the anode is placed behind it with the kathode in front rhythmical contractions are carried out by the sphincter. Ascending currents thus stimulate the central nervous system and descending depress it, and this law is seen to apply even in the case of but a single ganglion. Strange to say this interesting principle has attracted but little attention, although it appears to me to be of great significance. Since it is entirely distinct from the principles of electrotonus it is probably associated with the presence of nerve-cells within the central nervous system and also with the antero-posterior differentiation of the latter. The above results manifest not only a deviation from electrotonus but also from Pflüger's law. There are, however, other instances in which the anode and not the kathode

appears to be the stimulating electrode. Thus Loeb[1] found that in Amblystoma the skin-glands on the anodic side became active owing to stimulation of the central nervous system. Breuer[2] has obtained lateral flexion in the fish on the side towards the anode. I have studied the flexions of the tail of the crayfish at the make and break of the current and have found the anodic closing contraction to be the strongest of all. The kathodic opening contraction, though weaker than this, is stronger than the kathodic closing contraction. Breuer[3] has obtained analogous results in fish. The explanation of these facts and their bearing on Pflüger's law is not at all clear and further investigation appears most desirable.

In an attempt to analyse the galvanotropic behaviour of the crayfish it appeared desirable to determine the influence of the brain in this regard. Hermann[4] and Nagel[5] had experimented with decapitated animals, but the injury by this procedure is so great that the results cannot be considered trustworthy. In the writer's work the œsophageal commissures were cut after the manner described by Bethe[6], the dorsal route being selected by preference. Only large specimens were chosen as they are both easier to manipulate and resist shock much better than smaller ones. The animal is secured on its ventral surface to a board; one removes the carapace behind the rostrum, then goes down, cutting the anterior gastric muscles; the commissures which lie deeply are snipped with a pair of fine scissors. The opening is then carefully sealed by means of a mixture of beeswax and tallow.

After some hours the animal has recovered from the shock and then behaves in the manner described by Bethe[7]. There is the typical absence of spontaneous movements, the animal only going forwards when stimulated and then for but a short distance, with, however, the usual coordination of the legs. When not moving forwards the legs and pleopods perform pendulous movements. As is well known the forward movements of such animals depend on the integrity of the subœsophageal ganglion, which acts as a coordinating centre for the movements of progression. Under no circumstances whatever does the animal ever go backwards. No explanation of this latter peculiarity is attempted by Bethe; it may be, however, that the absence of backward

[1] *Pflüger's Arch.* LXV. p. 308. 1897.
[2] *Sitzungsb. d. Wiener Akad.* CXIV. p. 30. 1905.
[3] *Ibid.* p. 40.
[4] *Loc. cit.*
[5] *Pflüger's Arch.* LI. p. 624. 1892.
[6] *Ibid.* LXVIII. p. 459. 1897.
[7] [illegible] p. 4[illegible].

locomotion is due to the longitudinal giant-fibres, which run through the entire cord, having been sectioned.

How does such an animal behave under the influence of a constant current? In the first place as might be expected it never darts backward to the anode. But one kind of movement now obtains and that one forwards to the anode; it never goes to the kathode. The animal may advance to the anode immediately on closure of the current, at other times it appears to be in a condition of what Blasius and Schweizer have called galvanonarcosis, that is to say, it does not move forwards until the current is broken. This is particularly the case when one commences with a very weak current and gradually increases it. We have here probably to do with the paralysing influence of a descending current. There is thus the definite attraction to the anode just as occurs in the intact animal. It should be observed, however, that while the normal animal is somewhat capricious in its behaviour, occasionally going to the kathode, as mentioned above, this is never noticed after the commissures are cut. The behaviour of the appendages does not differ in any essential respect from that in the intact animal. When the anode is towards the head the legs and pleopods are inactive, while when the anode is towards the tail there is great excitement manifested by all the appendages as well as by the anus. Again we see the untenability of the position of Blasius and Schweizer, for the brain, which plays so important a part in their theory, is clearly, unnecessary for the production of the typical reactions. One more point deserves mention and that is the behaviour of the antennæ and antennules in such an animal: these invariably point towards the anode, *e.g.* if the anode is in front they point forwards, while if it is behind the antennules are bent underneath the thorax and the antennæ bent around to the sides. These reactions are also shown by the normal animal but to a minor degree; it is highly probable that they are inhibited in the intact animal by impulses which pass from behind forwards. It thus appears that the galvanotropic responses of the crayfish are intimately incorporated in its make-up and are a most fundamental property of the creature.

It is clear that the fundamental causative factor of galvanotropism is to be found in the peculiarities of the stimulus. Loeb and Budgett[1] had given an explanation from this standpoint but Statkewitsch[2] has lately thrown doubt upon its validity. I have tested the very plausible

[1] *loc. cit.*

[2] *Verworn's Ztschr. f. d. allgem. Physiol.* v. p. 529. 1905.

theory of Coehn and Barratt, mentioned in the introduction, and find that with minor modifications it applies to the crayfish. Their theory was proposed for *Paramœcium* which, as is well known, manifests a kathodic galvanotropism. Stated briefly their explanation is as follows: the membrane of the animal is supposed to be slightly more permeable for Cl than for Na ions. If, now, the animal is brought into a medium like tap-water the concentration of which is less than that of itself a slight diffusion of Cl ions will necessarily occur. The result will be that the animal will contain an excess of Na ions, or will bear a positive charge of electricity. On now subjecting the animal to a galvanic current it is definitely attracted to the kathode, and the cilia responding to the pull, it moves in this direction. A crucial test of the hypothesis consists in increasing the concentration of the medium in which the animals are observed. In this case the Cl ions outside should pass in, giving the animal a negative charge and it should hence move to the anode. Indeed this is found actually to occur: when *Paramœcia* are placed in N/10 NaCl solution, within the course of two or three minutes, during which time diffusion is taking place, the galvanotropic response is seen to be reversed and they move to the anode. The position of Coehn and Barratt has been lately criticised, but, in my opinion, not weakened by Bancroft[1].

In applying the theory of Coehn and Barratt to the crayfish certain peculiarities in the behaviour of the latter are to be taken into consideration. As already stated this form moves not to the kathode but to the anode; its reaction is just the reverse of that of *Paramœcium*. In order to explain this anodic galvanotropism one must suppose the animal to carry a negative charge of electricity, *e.g.* that its membranes are more permeable for Na than for Cl ions; the Na ions thus migrate out, leaving an excess of Cl ions. In order to produce reversal it will be necessary to place the animal in a salt solution, the osmotic pressure of which is greater than that of this same salt within the animal. The osmotic pressure of the crayfish has been determined by Frédericq[2] to equal that of a 1·3 % NaCl solution, corresponding to a depression of the freezing point of 0·8° C. This concentration represents, of course, that of all the salts contained in the animal. It was found possible to bring about reversal in the crayfish by strengths varying from N/5 to N/2 NaCl. N/2 NaCl equals 2·92 % NaCl and is thus considerably higher than the total concentration of the tissues of the animal, while

[1] *Univ. of Cal. Pub. Physiol.* III. p. 21. 1906. Also this *Journal*, XXXIV. p. 444. 1906.
[2] *Bull. de l'Acad. Roy. de Belg.* XXXV. p. 831. 1898.

N/5 NaCl, which equals 1·17 % NaCl, is somewhat less than this: this concentration of Na is, however, certainly higher than that of the Na contained in the tissues. The necessary conditions for obtaining the reversal are therefore fulfilled. Selected protocols are given below.

The procedure in making these experiments on reversal was as follows. Only freshly-collected, healthy animals were made use of, as it was found that those which were kept long in the laboratory became less sensitive and showed more irregularities than normal ones. The animals were first tested in tap-water to see whether their responses were normal. They were then placed each in a separate jar of N/5 NaCl and at once tested in this solution. Under these conditions they are still anodic. A control animal was kept in tap-water in every case and this was tested in N/5 NaCl at the same time as the other specimens. The reason for testing the control in the same solution as the other animals is obvious, as the conductivity and consequent stimulating action of the medium are thus the same. I may say that I have carried out many more experiments than those here reported and am convinced that a reversal can almost invariably be obtained. Very rarely I have failed, and in these cases the failure was due to the poor condition of the animals used. I believe that in fresh, healthy specimens the reversal will occur in every instance. Different strengths of current were used as measured by the galvanometer.

From the above results it is clear that the law established by Coehn and Barratt in the case of *Paramœcium* holds also in the crayfish. We notice, however, certain differences between the two forms. For the reversal to be brought about several hours, instead of minutes, are required. One may attribute this slowness to the greater impermeability of the membranes of the crayfish. It is not possible to state with certainty through which membranes the migration of ions takes place; but it is not improbable that part of the exchange occurs through the gills. One can see in this way how the process would be necessarily a slow one. Frédericq[1] also is of this opinion.

The positions of the limbs and abdomen constituting the orientations are found to be unaltered after the reversal. These are therefore not the cause of the reaction, but merely represent a parallel series of phenomena, depending as pointed out above partly on stimulation of the central nervous system and partly also on stimulation of the peripheral nerves. Moreover the relative irritabilities of the muscles, on which these depend, are found to be unchanged. It must also be affirmed that while the action of the constant current on the central nervous system, in producing stimulation in the ascending, and depression in the descending, direction as observed first by Hermann, is

[1] *Bull. de l'Acad. Roy. de Belg.* xxxv. p. 68. 1901.

No. of Exp.	Date	Time	No. of animals	Anode	Kathode	Behaviour of Control	Currents	Remarks
I.	July 20, '06	10 a.m.	4	4	0	anodic	33·78—64·48	
$\frac{N}{5}$ NaCl		2.45 p.m.		1	3	,,	,,	Animals partly reversed.
		5.15 p.m.		0	4	,,	,,	Animals completely reversed.
		6.30 p.m.		0	4	,,	,,	,, ,, ,,
	July 21, '06	9.05 p.m.		0	4	,,	,,	,, ,, ,,
II.	July 28, '06	10.15 a.m.	2	2	0	,,	21·48—56·48	
$\frac{N}{5}$ NaCl		12.15 p.m.		2	0	,,	,,	
		2.10 p.m.				,,	,,	Animals go to anode and kathode.
		4.50 p.m.		0	2	,,	,,	Animals completely reversed.
		6 p.m.		0	2	,,	,,	,, ,, ,,
	July 29, '06	11 a.m.		0	2	,,	,,	,, ,, ,,
III.	Aug. 11, '06	10.23 a.m.	1	1	0	,,	73·78—14·18	
$\frac{N}{2}$ NaCl		11.40 a.m.		1	0	,,	,,	
		12.20 p.m.		1	0	,,	,,	
		2.45 p.m.				,,	,,	Animal goes to both anode and kathode.
		3.40 p.m.				,,	,,	,, ,, ,, ,, ,,
		4.25 p.m.		0	1	,,	,,	Animal entirely reversed.
		5.50 p.m.		0	1	,,	,,	,, ,, ,,
		7 p.m.		0	1	,,	,,	,, ,, ,,
	Aug. 12, '06	9.30 a.m.		0	1	,,	,,	,, ,, ,,
IV.	Sept 28, '06	10.55 a.m.	4	4	0	,,	27·68—67·48	
$\frac{N}{5}$ NaCl		2 p.m.				,,	,,	Animals go to both anode and kathode.
		4 p.m.		0	4	,,	,,	Animals completely reversed.
		5.30 p.m.		0	4	,,	,,	,, ,, ,,
	Sept. 29, '06	9 a.m.		0	4	,,	,,	,, ,, ,,

sufficient to lead mechanically to directive orientations there is in these nothing *per se* to induce a definite galvanotropic attraction. That such exists, however, cannot be doubted by anyone who has watched the crayfish under the influence of the constant current; no matter what its attitude at the moment of closure, it is drawn to the anode, as it were, by an invisible force.

The attraction of the animal thus depends upon the carrying of an electrical charge and this in turn is associated with a peculiarity on the part of its membranes. It is highly probable that in those forms which do not exhibit any attraction one way or the other the membranes are impermeable. As is well known the teleosts are not attracted to either the anode or the kathode, and here we might assume the membranes to be impermeable. Other facts lend probability to such a view. By cryoscopic determination Bottazzi[1] has found the concentration of the blood of marine teleosts to be one-half that of sea-water. Garrey[2] has found that their osmotic pressure cannot be altered by placing them in more dilute or more concentrated media, unless the membranes are previously injured. He therefore concludes that their integument and gills are impermeable. Here then we have the explanation of the absence of galvanotropic attraction among these forms.

Summary.

1. The orientations of the crayfish, when under the influence of the constant current, are produced by the combined effect of stimulation of the central nervous system and of the peripheral nerves. The impulses sent out are of such a strength that one group of muscles is caused to contract, while the antagonistic group is at the same time inhibited.

2. The ascending current leads to stimulation of the central nervous system, while the descending current causes depression of its irritability.

3. The movement of the animal forwards, but not backwards, to the anode is preserved after section of the œsophageal commissures.

4. The theory of Coehn and Barratt that the attraction of *Paramœcia* depends on their carrying electrical charges applies with modifications in the case of the crayfish.

5. It is probable that the impermeability of the membranes of such forms as teleosts determines the absence of any galvanotropic attraction among them.

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[1] Bottazzi. *Arch. Ital. de Biol.* xxviii. p. 61. 1897.
[2] *Biol. Bull. of Mar. Biol. Lab. Woods Holl.* viii. p. 266. 1905.